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PAPER

The influence of two different breeding systems on quality and clotting properties of milk from dairy buffaloes reared in Sicily (Italy)

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Abstract

This study focuses on nine buffalo farms (a total of 692 buffaloes), five intensive herds (161 lactating buffalo cows) and four semi-extensive herds (111 lactating buffalo cows), which are the most common in Sicily. The average milk and cheese yield, fat, protein and lactose contents were very similar between the two breeding systems. Milk casein was higher ($P=0.04$) in intensive herds than in semi-extensive herds (4.07% vs 3.51%), whereas the urea content was higher in semi-extensive herds (40.68 mg/dL vs 37.50 mg/dL), yet within the normal range for buffalo milk. As regards milk pH and titration acidity ($^{\circ}\text{SH}$), no difference was observed between the two breeding systems. Milk produced by the intensive herds scored the best clotting ability with shorter curd firming time (k_{20} 3.08 min) and greater curd firmness (a_{30} 32.69 mm). Milk produced by the semi-extensive herds, instead, was characterized by a longer curd firming time (k_{20} 5.26 min) and lower curd firmness (a_{30} 27.33 mm). This milk from buffaloes bred in Sicily (Italy) also showed a good chemical composition and clotting ability in particular when produced by the intensive herds.

Introduction

The buffalo population in the Mediterranean area, which has typical climatic and cultural conditions and includes Europe

and the countries of the Near East covered by the FAO Inter-Regional Cooperative Research Network on Buffalo, accounts for about 5.5 million head, 3.4% of the world buffalo population, that currently includes about 174 million head (Tiezzi *et al.*, 2009; Bittante, 2011).

In Italy the increasing demand for buffalo *Mozzarella* cheese results both from the national and international markets of Protected Denomination of Origin (PDO) *Mozzarella*, known as *Mozzarella di Bufala Campana*, and the milk quotas introduced by the European Union (EU) to prevent bovine milk overproduction, which led to an increase in the buffalo population to 365,086 head (Banca Dati Nazionale, 2014). *Mozzarella di Bufala Campana* is produced according to the guidelines approved by the EU (EU, 1996), which set out specific requirements in terms of origin of the buffalo milk to be used and processing conditions (Brescia *et al.*, 2005). In Italy this increase appears to be quite interesting also in view of the decrease of other species over the last 50 years, particularly of cattle, dairy cows, sheep and horses.

The challenges facing the Italian dairy cow sector has mainly been caused by the milk quotas imposed by the EU, because of the structural and smaller size of farms in comparison with those of northern European countries, Bovine Spongiform Encephalitis and Blue Tongue Disease and a reduction in cows' reproductive life, which now does not exceed 2.5-3 calvings. The ovine livestock is also penalized by the new health regulations implemented in the EU, insufficient pasture availability, Blue Tongue Disease, milk industry problems and changes in the demand of consumers who tend to prefer fresh and soft cheeses (such as *Mozzarella*) over dry cheeses (such as pecorino) (Borghese, 2005). The recent intensification of buffalo rearing techniques has exposed them to a rapidly changing environment that imposes physical and psychological stressors so far unknown to this species. Machine milking presents both physical (poor maintenance of the machine) and psychological factors (negative behaviour of the stock person and calf separation) which may interfere with milk ejection (Borghese *et al.*, 2007). As reported by Thomas *et al.* (2003) buffaloes are known to be difficult to milk. In order to overcome these problems, 65% of the farms used concentrate feeding during pre-stimulation to improve milk let-down, while in 13% oxytocin was injected to induce milk ejection. Human-animal interactions (De Rosa *et al.*, 2005) are recognized to have an impact on productivity, behaviour and welfare, particularly in dairy farms, where the milking process

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involves close interaction with a human operator.

Buffalo milk, which is characterized by a higher fat (8.3-8.6%) and protein (4.5-4.8%) content compared with cow's milk, yields between 2100 and 2300 kg of cheese over 270 days (Zullo, 2007; Rosati and Van Vleck, 2002). The buffalo population in Sicily (Italy) includes over 1000 buffaloes (1336 head) reared in 19 farms (BDN, 2014). Their milk is processed to make typical cheeses and is sometimes mixed with cow's milk. This study was aimed to provide an update on the situation of buffalo breeders in Sicily and shed more light on their problems. It was also intended to collect some general information about buffalo management practices, breeding systems and milk traits in Sicily.

Materials and methods

Animals and breeding

The study was carried out from March to June 2013 on 271 multiparous lactating buffaloes out of a total of 692 buffaloes belonging to the Mediterranean breed reared in 9 buffalo farms, 5 intensive herds (on average, 161 lactating buffalo cows) and 4 semi-extensive

herds (on average, 111 lactating buffalo cows), located in Sicily. In the intensive breeding system, all buffaloes were kept in loose housing systems, without pasture, and fed with a total mixed ratio (concentrate 9 kg/head/d and vetch hay 10.5 kg/head/d), distributed twice a day. In the semi-extensive system, the facilities were split between an indoor area and an outdoor area. In the indoor area, covered with straw

bedding, animals were fed with concentrate (6 kg/head/d) and meadow hay ad libitum. In the adjacent outdoor area (on average, space allowance of 500 m² per head) animals had free access to green natural pasture (from 8 a.m. to 6 p.m.) with vetch, ryegrass and barley in equal parts. Animals were treated with Ivomec® against ecto and endo-parasites.

Behavioural observations: oxytocin administration

For to evaluate the behavioural responses of buffalo of stock persons during machine milking (Cavallina *et al.*, 2008) was recorded the number of buffaloes to which was oxytocin at milking administered.

Table 1. Chemical composition of feeds in intensive and semi-extensive buffalo farms.

	Breeding system				
	Intensive		Semi-extensive		
	Complete feed	Pellet Vetch hay	Complete feed	Meal	Meadow hay
Dry matter, %	88.80	87.50	88.59		86.00
Moisture, %	11.20	12.50	11.41		14.00
Crude protein, % DM	20.06	18.60	14.11		11.09
Crude fat, % DM	4.15	2.67	3.43		2.50
Crude fibre, % DM	19.40	24.10	5.15		27.49
Ash, % DM	4.55	10.00	7.91		9.00
Neutral detergent fibre, % DM	19.40	42.72	17.47		52.19
Acid detergent fibre, % DM	11.25	29.37	6.30		27.86
Acid detergent lignin, % DM	2.77	7.12	1.30		8.50
NEL ^o , on DM	1.08	0.52	1.09		0.65

^oNet Energy Lactation (French units) expressed as FUL Feed Unit for Lactation: FUL, NEL (kcal)/1700(kcal).

Table 2. Milk traits in intensive and semi-extensive buffalo farms (mean ± SE).

	Breeding system		P-value
	Intensive	Semi-extensive	
Herd, n	5	4	
Lactating buffalo cows, n	161	111	
Oxytocin administration, head	17	23	0.05
Milk yield, kg/head/d	7.34±1.04	7.50±3.11	0.46
Cheese yield, %	27.20±2.17	28.75±1.50	0.13
Fat, %	8.78±2.26	9.37±4.20	0.40
Protein, %	4.61±0.50	4.29±0.10	0.12
Casein, %	4.07±0.37	3.51±0.42	0.04
Lactose, %	4.72±0.27	4.54±0.60	0.27
Total solids, %	18.39±2.16	18.34±3.14	0.49
Non fat solids	10.53±0.36	9.87±0.57	0.04
Urea, mg/dL	37.50±13.89	40.68±23.89	0.40
Cryoscopy index, m°C	0.53±0.02	0.54±0.02	0.24

Table 3. Clotting properties of milk in intensive and semi-extensive buffalo farms (mean ± SE).

	Breeding system		P-value
	Intensive	Semi-extensive	
Herd, n	5	4	
Lactating buffalo cows, n	161	111	
pH	6.7±0.03	6.6±0.05	0.55
Titration acidity, °SH/100 mL	17.72±3.76	17.65±4.32	0.49
Clotting time (r), min	21.30±2.01	19.43±1.13	0.002
Curd firming time (k ₂₀), min	3.08±0.56	5.26±1.43	<0.001
Curd firmness (a ₃₀), min	32.69±6.30	27.33±5.09	0.007

Samplings

For each herd, bulk milk samples (morning and evening milkings) were collected every two weeks along with feed samples, while also milk production was recorded. The milk samples of approximately 250 mL were collected into high-density polyethylene Azlon bottles (Barloworld Scientific, Riozzo di Cerro al Lambro, MI, Italy), stored at 4°C and analysed within 24 h. Milk yield, cheese yield data and feed samples were collected by each farmer.

Laboratory analysis

Milk samples were analysed to measure fat, protein, casein, lactose, total solids, non-fat solids, urea, cryoscopy index and titration acidity (Soxhlet-Henkel/°SH) using Fourier Transform InfraRed (Milkoscan FT2, Foss Electric, Sweden), calibrated with appropriate buffalo standards. Moreover, rennet clotting time (t_r , in min), curd firming time (k_{20} , in min) and curd firmness, which was measured 30 minutes after the addition of rennet (a_{30} , in mm), were determined with a Formagraph (Foss Electric, Hillerød, Denmark) in 10 mL of milk at 35°C with a 200 μ L dose of Hansen Standard rennet (diluted 1:100) according to the method developed by the (ASPA) (Animal Science and Production Association) (1995). The Association of Analytical Chemists (AOAC) official methods (2007) were used to determine the chemical composition of feeds (Table 1).

Statistical analysis

The statistical analysis of milk traits was performed using a one-way analysis of variance with repeated measures of the GLM procedure by SAS (2001), considering as variable the breeding systems (intensive *vs* semi-extensive). Means were separated by a Least Significant Difference test when a significant treatment ($P < 0.05$) was observed.

Results and discussion

The frequency of oxytocin administration (Table 2) was significantly higher ($P < 0.05$) in buffalo cows bred with the semi-extensive system (intensive 17 head corresponding to 10.5% *vs* semi-extensive 23 head corresponding to 21.6%). As reported by Borghese *et al.* (2007), in Italy, it is normal to find at least 15-20% of buffalo cows receiving an oxytocin injection before milking in each farm. In a study carried out in Italy on 17 buffalo farms and 1030 Mediterranean buffaloes (Saltalamacchia *et al.*, 2005), 13% of all lactating buffaloes were

treated with oxytocin before milking. However, 24% of primiparous buffalo cows received oxytocin injections. Therefore authors suggested that the difficulties in milk let-down were more prominent in primiparous than multiparous buffaloes. The results of our study confirmed that the behaviour of stock persons affects buffalo behaviour at milking, which is also impacted by the use of oxytocin injections.

The milk traits of the two breeding systems are reported in Table 2 and are within the normal range for this species bred in Italy (Bartocci *et al.*, 2002; Bovera *et al.*, 2002; Catillo *et al.*, 2002; Calabrò *et al.*, 2003; Di Francia *et al.*, 2003; Zicarelli, 2004; Tudisco *et al.*, 2010). The average milk and cheese yield, fat, protein, lactose percentages and urea content were very similar between the different feeding systems used in the two breeding systems (Table 2). The casein percentage in the milk of buffaloes reared with the intensive breeding system was higher ($P = 0.04$) than in the semi-extensive system (4.07% *vs* 3.51%). The urea content was in line with the observations of Frand *et al.* (2003) in cattle breeding both in pasture and stalls and of Campanile *et al.* (1998) in buffalo cows fed diets with a different protein content.

No difference was observed between the two breeding systems as regards milk pH and °SH (Table 3). Milk reported the best clotting ability in buffaloes reared in the intensive breeding system and was characterized by greater curd firmness (a_{30} 32.69 mm) and shorter curd firming time (k_{20} 3.08 min). This milk, which generally produces good-quality cheese, is characterized by shorter curd firming time and good curd firmness (Bartocci *et al.*, 2002; Cipolat Gotet *et al.*, 2010). On the contrary, milk from the semi-extensive herd was characterized by a longer curd firming time (k_{20} 5.26 min) and lower curd firmness (a_{30} 27.33 mm). These results could be related to the lower casein content and higher urea content in the semi-extensive breeding system. The k_{20} value, according to Potena *et al.* (2007), is however inversely correlated to a_{30} .

Conclusions

This study demonstrates that Mediterranean buffaloes reared in Sicily show a good productivity level and produce good quality milk with a good clotting ability. The feeding system used in the intensive herd showed the best milk quality characteristics. In order to enhance the technological performance of buffalo milk, others studies would be

necessary to improve feeds and herd management practices.

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